REMARKS/ARGUMENTS

Petition is hereby made under the provisions of 37 CFR 1.136(a) for an extension of three months of the period for response to the Office Action. The enclosed cheque includes the prescribed fee.

The Examiner withdrew claim 9 (insofar as the claim covers species other than the elected Hia species) and claims 14 to 43 from further consideration as being directed to a non-elected invention or species, there being no elected generic or linking claim. Claims 14 to 43 have been deleted from the application, such deletion being made without prejudice to applicants right to file one or more continuation or divisional applications directed thereto.

Claim 9 has been amended to delete reference to a non-elected species. Having regard thereto, it is requested that there be rejoinder of claim 9 and claim 9, specifically directed to the elected species Hia, be examined with the remainder of the pending claims.

The Examiner noted that the IDS filed July 26, 2001 failed, in part, to comply with the provisions of 37 CFR 1.97, 1.98 and MPEP 609 because it lacks copies of several of the non-patent literature references were not enclosed, but further indicated that they were to follow. It is regretted that the missing copies were not forwarded to the PTO.

To remedy this oversight, enclosed herewith is a further PTO-1449 listing these missing references and enclosing copies thereof. In addition, this PTO-1449 includes a listing of references cited in the corresponding PCT filing to the extent not already cited in the IDS previously submitted, and copies of this prior art is enclosed. The enclosed cheque includes the prescribed fee for submission of the PTO-1449 at this stage of prosecution.

The Examiner indicated that the proposed drawing corrections were acceptable and that corrected drawings were required to be submitted in response to the Office Action. The corrected drawings are enclosed.

The Examiner rejected claims 1 to 3 under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 7 and 11 of US Patent No. 5,506,139.

Claims 1 to 3 are directed to an expression vector which comprises a nucleic acid molecule encoding a non-proteolytic analog of a Hin47 protein of a strain of *Haemophilus* including a portion thereof encoding the leader sequence for the non-proteolytic analog and a promoter operatively connected to the nucleic acid molecule to direct expression of the non-proteolytic analog of the Hin47 protein having the leader sequence.

The preparation of the natural non-proteolytic analog of Hin47 is described in USP 5,506,139. The protein, lacking the leader sequence, was produced at 40 to 50% of total protein in a soluble form. Claim 7 of the US Patent claims an isolated and purified nucleic acid molecule comprising a mutant *Haemophilus influenzae* hin47 gene coding an analog of *Haemophilus influenzae* protein having a decreased protease activity which is less than about 10% of that of natural Hin47 protein having specific codons deleted or replaced by a codon encoding a different amino acid, while claim 11 recites a specific codon mutation.

As the Examiner correctly points out, the claims of USP 5,506,139 in question do not recite a leader sequence or a promoter, as required by claims 1 to 3 of this application. The Examiner considered that these differences did not make the currently pending claims patentably distinct from claims 7 and 11 of USP 5,506,139.

The Examiner noted that the promoter is taught in the patent, which is agreed. The Examiner indicated that the presence or absence of the leader sequence is not discussed in the patent, but that the sequence is present in the Hin47 sequence shown in Figure 3 of the patent, identifying the Hin47 sequence as SEQ ID No: 2. SEQ ID No: 2 is the deduced amino acid sequence while SEQ ID No: 1 is the full nucleotide sequence for the unmodified Hin47 gene.

Claims 1 to 3 are directed to an expression vector, an entity not claimed in USP 5,506,139. The expression vector contains a mutant Hin47 gene with its leader sequence and expression of protein with the leader sequence. It is submitted that such

claimed subject matter is not an obviousness-type double patenting of claims 7 and 11 of USP 5,506,139. While, as the Examiner notes, the rejection maybe overcome by the filing of a Terminal Disclaimer, such disclaimer is not required as the claims are patentably distinct.

Accordingly, the rejection of claims 1 to 3 under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 7 and 11 of US Patent No. 5,506,139, should be withdrawn.

The Examiner objected to the disclosure in that the specification in several locations refers to Patent Application No. 09/268,347, when, in fact, the application has been patented as US Patent No. 6,335,182. The references to Patent Application No. 09/268,347 have now been replaced by references to the corresponding US Patent No. 6,335,182.

The Examiner rejected claims 1, 5, 6, 9 to 11 and 14 under 35 USC 112, first paragraph, on the basis that the specification, which enabling for an expression vector encoding a non-proteolytic analog created by the substitution or deletion of one or more of the amino acid residence 91, 121 and 197, does not reasonably provide enablement for any non-proteolytic Hin47 analog.

The specification identifies the analog as non-proteolytic and provides cross-reference to US Patent No. 5,506,139. The reference describes the manner of providing the analog, namely identifying amino acids contributing for protease activity and deleting or modifying the amino acid. The patent describes the manner of determining which amino acids contribute to protease activity. The disclosure of USP 5,506,139 was considered sufficient to support a claim to an isolated and purified analog of Hin47 protein having a decrease protease activity which is less than about 10% of that of natural Hin47 protein. In this regard, the Examiner's attention is directed to claim 1 of USP 5,939,297.

Accordingly, the rejection of claims 1 and 5, and dependent claims 6, 9 to 11 and 14, under 35 USC 112, first paragraph, should be withdrawn.

The Examiner rejected claims 1, 2, 5, 6 and 7 under 35 USC 112, first paragraph, on the basis that the specification, while enabling for a non-proteolytic

Hin47 analog wherein the analog include substitution or deletion of at least one of the amino acid resides 91, 121 or 197, does not reasonably provide enablement to the analog with the substitution or deletion of amino acid 195, 196 or 198 to 201.

While the applicants have not specifically demonstrated that substitution or deletion of any of those amino acids leads to loss of proteolytic activity, there is no reason to suppose that mutation at such sites would not result in loss of protease activity. As explained in the cited Loosmore et al patent (USP 5,506,139), col. 5, lines 55 to 61, there is an art recognized consensus sequence of serine proteases in which the active residue in serine. The three-dimensional structure of the molecule in this region in such that either direct mutation of serine or one of the adjacent amino acids removes the proteolytic activity, since the serine no longer is exposed.

Accordingly, it is submitted that claims 1, 2, 5, 6 and 7 are fully enabled and hence the rejection thereof under 35 USC 112, first paragraph, should be withdrawn.

The Examiner rejected claims 4, 12 and 13 under 35 USC 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

With respect thereto, claims 4, 12 and 13 have been amended to refer specifically to the claimed plasmids. It is submitted that claims 4, 12 and 13 can no longer be considered indefinite and hence the rejection thereof under 35 USC 112, first paragraph, should be withdrawn.

The Examiner rejected claims 1 to 3 under 35 USC 102(b) as unpatentable over Loosmore et al (1998 Infec. Immun. Article).

Claim 1 defines an expression vector comprising:

- a nucleic acid molecule encoding a non-proteolytic analog of a Hin47 protein of a strain of *Haemophilus* including a protein thereof encoding the leader sequence for the non-proteolytic analog

- a promoter operatively connected to the nucleic acid molecule to direct expression of the non-proteolytic analog of a Hin47 protein having the leader sequence.

Such an expression vector is not described in the 1998 Article. The Examiner refers to page 900, col. 1, as indicated under the heading "Expression of recombinant htrA and generation of mutant protein", the smallest clone containing all of the *htrA* gene (i.e. the gene encoding Hin47) was used to construct expression plasmids. This clone does not contain any promoter and itself is incapable of expressing the protein. The Thr at position 27 was assumed to represent the start of the native protein. As stated:

> "Oligonucleotides were synthesized to encode the N-terminus of the mature HtrA up to the *Bsp* MI site Figure 3)". (emphasis added)

Figure 3 shows the oligonucleotides which encode the N-terminus of the mature HtrA protein. If the amino acids sequence depicted in Figure 3 is compared to the amino acid sequence shown in Figure 4 (a sequence alignment of HtrA proteins from different *H. influenzae* strains), it will be seen that, apart from the leading methionine, present to ensure expression of the mature protein, the amino acids are the tyrosine at aa27 and the next twenty-two amino acids of the mature protein sequence shown in Figure 4.

Thus, in the 1998 paper, the expression vectors are constructed to express the Hin47 protein or its non-proteolytic analog without the leader sequence. There is no construction of a plasmid expression vector in which there is a nucleic acid molecule encoding a non-proteolytic analog, including a portion thereof encoding the leader sequence for the non-proteolytic analog, as required by claim 1.

Accordingly, claims 1 to 3 are not anticipated by the cited prior art and hence the rejection thereof under 35 USC 102(b) as being anticipated by the 1998 article, should be withdrawn.

The Examiner rejected claim 4 under 35 USC 102(b) as being anticipated by the Loosmore et al '139 patent.

This rejection appears to be based on the term "having the identifying characteristics" previously employed. As noted above, this language has been removed from claim 4. It is submitted that '139 patent does not disclose plasmid JB-3120-2 as seen in Figure 1A.

Accordingly, it is submitted that claim 4 is not anticipated by the '139 patent and hence the rejection thereof under 35 USC 102(b) should be withdrawn.

The Examiner rejected claim 4 under 35 USC 103(a) as being unpatentable over the '139 patent in the light of USP 6,361,969 to Galeotti (the Galeotti patent) and Recombinant DNA, 2nd edition by Watson et al.

The '139 patent, is discussed above and does not describe plasmid JB-3120-2. While the Recombinant DNA reference teach that plasmids are practical both for gene cloning and expression, it is not seen how this adds to the teaching of the '139 patent in relation to the specific plasmid claimed in claim 4.

Similarly, while the Galeotti patent refers generally to expression of a fusion protein comprised of a leader sequence fragment, it is not seen how this adds to the teaching of the '139 patent in relation to the specific plasmid claimed in claim 4.

Accordingly, it is submitted that claim 4 is patentable over the applied prior art and hence the rejection thereof under 35 USC 103(a) as being unpatentable over the '139 patent in light of the Galeotti patent and the Recombinant DNA, should be withdrawn.

The Examiner rejected claims 5 to 8 under 35 USC 103(a) as being unpatentable over Bass et al J. Bacteriology in view of the 1998 article and US Patent No. 5,474,914 to Spaete.

Claims 5 to 8 define an expression vector for expression of a recombinant protein in the host cell. This expression vector comprises several elements:

- a nucleic acid molecule encoding a non-proteolytic analog of a *Haemophilus* Hin47 protein

- at least one additional nucleic acid molecule encoding the recombinant protein

- at least one regulatory element operatively connected to the first nucleic acid molecule and to the at least one additional nucleic acid molecule to effect expression of at least the recombinant protein in the cell.

As the Examiner states, on page 1157, the Bass et al article indicates that the HtrA family of proteins may act as chaperones by binding perisplasmic proteins that have just been secreted or those that are denatured under stressful conditions. It is agreed with the Examiner that the Hin47 protein of *Haemophilus* is an HtrA protein. This reference is silent as to any non-proteolytic analogs of Hin47 and their potential utility as chaperone proteins. In fact, the Examiner states:

> "... Bass does not teach a non-proteolytic Hin47, or the co-expression of Hin47 (with or without a leader sequence) with another protein to facilitate the secretion of itself and the other protein."

The contents of the 1998 article have been discussed above. While this reference teaches the non-proteolytic analog of the Hin47 protein of *Haemophilus*, the article is silent with respect to the potential use of the analog as a chaperone protein. As the Examiner states:

> "... the article does not teach the combination of Hin47 with another protein."

The Examiner already stated that the Bass reference does not teach co-expression of Hin47 with another protein and that the 1998 article does not teach the combination of non-proteolytic Hin47 with another protein. The Examiner justifies his stated rejection by saying:

> "As Hin47 is no longer lytic, there is no longer a concern as to whether or not the other protein will be degraded or protected.

However, there is no manner of determining from the teachings of the two references whether or not a non-proteolytic Hin47 protein, in fact, will function as a chaperone protein in the absence of the protease activity.

The Spaete reference describes a method of recombinantly producing a protein by coexpressing in a host cell a first gene encoding the protein with a second gene encoding an escort under conditions whereby the protein is secreted from the host cell. Applicants are not claiming a general procedure for expressing secreted proteins, but a specific plasmid containing a nucleotide sequence encoding a non-proteolytic analog of Hin47 protein of *Haemophilus*.

While the reference refers to and describes that any desired protein can be produced using a compatible escort, the Spaete reference is specifically concerned with secreting truncated cytomegalovirus glycoprotein using suitable fibroblast growth factor receptor as the chaperone and the claims of the patent are so limited. Since the claims of Spaete are so limited, it is clear that the Spaete reference lacks enablement and written description to support generic claims. In common with Galeotti discussed above (the Galeotti patent and the Spaete patent have a common assignee), the Spaete patent refers to secretion of chimeric DNA molecules that encode fusion proteins comprised of a leader sequence fragment and a foreign protein. The fact that the DNA is described as "chimeric" (col. 21, line 46) suggests that the leader sequence is a heterologous rather than a homologous leader sequence.

As the Examiner admits:

> "The reference does not specifically teach the use of a Hin47 analog as a chaperon protein."

However, the Examiner asserts that:

> "..... one or ordinary skill in the art would have known from the to [sic-two] references above that Hin47 could be used in the disclosed vector."

The relevance of the Bass reference and the 1998 article and their combined teachings have been discussed above. As set forth above, the combination of the two reference does not teach that a non-proteolytic analog of Hin47 may be used as a chaperon protein to other proteins. Since the Spaete reference does not describe the use of a non-proteolytic Hin47 analog as a chaperone protein, on the Examiner's own admission, the reference does not remedy the defects of the combination of Bass and the 1998 article.

Accordingly, it is submitted that claims 5 to 8 are patentable over the applied combination of prior art and hence the rejection thereof under 35 USC 103(a) as being unpatentable over Bass, the 1998 article and the Spaete patent, should be withdrawn.

The Examiner rejected claims 9 to 11 under 35 USC 103(a) as being unpatentable over Bass in view of the 1998 article and the Spaete patent and further in view of Barenkamp and St. Geme III and USP 6,335,182.

Claim 9 is directed to the at least one additional nucleic acid molecule encoding a Hia protein of a strain of *Haemophilus influenzae*. Claim 10 is directed to N-terminal truncation of the encoded Hia protein while claim 11 recites two specific terminal truncations.

The combined teachings of Bass, the 1998 article and the Spaete patent have been discussed above. The Examiner summarizes his view of these teachings as follows:

> "These references therefore show that it would have been obvious to one of ordinary skill in the art to make an expression vector encoding for a Hin47 non-proteolytic analog and another protein to be recombinantly expressed."

As noted above, it is the applicants view that such is not the case, in that the combination of references does not disclose or suggest that the non-proteolytic analog acts as a chaperone and hence there is no motivation to construct a vector as defined in claim 5.

As the Examiner indicates:

> "..... the references do not teach that the other protein to be expressed is the *Haemophilus influenzae* Hia protein."

The Examiner relies on the secondary references to attempt to remedy this defect as it applies to claims 9 to 11. It is agreed with the Examiner that Barenkamp describes the existence of the Hia protein of *Haemophilus influenzae*, molecular cloning of the gene encoding the protein and construction of expression vectors.

The Examiner asserts that:

> "Barenkamp teaches that the Hia protein may also be used as a vaccine against *Haemophilus influenzae*, especially in combination with other antigens. Pp. 1220-21"

what is actually stated by Barenkamp is:

> ".... its immunogenicity and role as an adhesin suggests its potential role as a vaccine candidate. If combined with representative HMW1/HMW2-like protein in a vaccine formulation could be envisioned that would be protective against most or all non-typeable *H. influenzae*." (emphasis added)

This is far short of the Examiner's statement in several respects.

The Examiner asserts:

> "Thus, it would have been obvious to one of ordinary skill in the art to combine Hin47 analog and Hia to make a multiantigenic vaccine for *Haemophilus influenzae*. Knowing that the two proteins could be used together, it would have been obvious to one of ordinary skill in the art to make an expression system that produces both antigens."

First of all, Barenkamp contains no indication of combining Hin47 with Hia in a multiantigen vaccine. The only suggestion in Barenkamp is that there is envisioned the possibility of combining Hia with HMW1/HMW2 proteins of *Haemophilus influenzae* in a vaccine formulation. There is no mention at all of Hin47.

The teachings of Barenkamp do not make it obvious to construct a vector as defined in claim 9. As already noted, the basic combination of references is defective in that there is no suggestion therein that a non-proteolytic analog of Hin47 may be employed as a chaperone protein and, therefore, no suggestion to provide a vector as defined in claim 5.

In addition, there is no information at all provided by the combination of Bass, the 1998 article and the Spaete patent in combination with Barenkamp that a non-proteolytic analog of Hin47 protein of *Haemophilus influenzae* may act as a chaperone for the specific Hia protein described by Barenkamp.

As the Examiner observes, the '182 patent describes the recombinant production of N-terminally truncation forms of the Hia protein of *Haemophilus influenzae*, and specifically the V38 truncation specifically claimed in claim 11, and that such materials are useful as antigens in immunogenic compositions (col. 3, lines 45 to 47 to which the Examiner refers does not use the term "vaccine antigen" used by the Examiner).

However, this reference does not remedy the basic defects of the combination of Bass, the 1998 article, the Spaete patent and Barenkamp described above. Accordingly, it is submitted that claims 9 to 11 are patentable over the applied art and hence the rejection thereof under 35 USC 103(a) as being unpatentable over the combination of Bass, the 1998 article, the Spaete patent, Barenkamp and the '182 patent, should be withdrawn.

The Examiner rejected claims 12 under 35 USC 103(a) as being unpatentable over Bass, the 1998 article, and the Spaete patent and further in view of Barenkamp, the '182 patent and Recombinant DNA. In the body of the rejection, the Examiner refers to the '139 patent, but does not include it in this rejection.

Claim 12 is directed specifically to plasmid DS-2342-2-2 as seen in Figure 5. This plasmid contains *T7H91Ahin47* and *T7V38hia* gene cassettes with the *H91Ahin47* gene encoding the mature protein. It is noted that claim 13 is directed to a similar plasmid except that the *H91Ahin47* gene encodes the protein with its leader sequence. There is no prior art rejection of claim 13.

The rejection arises, in part, apparently, from the prior use of terminology "plasmid with the identifying characteristics", which now has been deleted. The teachings of Bass, the 1998 article, the Spaete patent, Barenkamp and the '182 patent have been described above. The combined teachings, admittedly by the inclusion of additional references, does not disclose or suggest any plasmid containing the two cassettes used in the plasmid of Figure 5.

It is submitted that the Recombinant DNA article, with or without the '139 patent referred to in the body of the discussion but not in the rejection, does not remedy these defects and that there is no disclosure or suggestion in the prior art to provide plasmid DS-2342-2-2 specifically claimed in claim 12. These additional references were discussed above in relation to claim 4.

The '139 patent describes the recombinant production of a non-proteoltyic analog of Hin47 protein of *Haemophilus influenzae*. While the Recombinant DNA reference teaches the plasmids are practical both for gene cloning and expression, it is not seen how these teachings add to the combined

teachings of the basic combination of references sufficient to render obvious the specific plasmid defined in claim 12.

Having regard thereto, it is submitted that claim 12 is patentable over the applied combination of prior art and hence the rejection of claim 12 under 35 USC 103(a) as being unpatentable over Bass in view of the 1998 article, and the Spaete patent, and further in view of Barenkamp, the '182 patent and Recombinant DNA (with or without the '139 patent), should be withdrawn.

Attached hereto is a marked-up version of the changes made to the specification and claims by the current amendment. The attached page is captioned "**Version with markings to show changes made**."

It is believed that this application is now in condition for allowance and early and favourable consideration and allowance are respectfully solicited.

Respectfully submitted,

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VERSION WITH MARKINGS TO SHOW CHANGES MADE

In the Specification:

Please replace the paragraph beginning at page 3, line 9, with the following rewritten paragraph:

"A second family of high molecular weight adhesion proteins has been identified in about 25% of NTHI and in encapsulated *H. influenzae* strains (refs. 11, 12, 13). The NTHi member of this second family is termed *Haemophilus influenzae* adhesin or Hia, and the homologous protein found in encapsulated strains is termed *Haemophilus influenzae* surface fibril protein or Hsf. The *hia* gene was originally cloned from an expression library using convalescent sera from an otitis media patient, which indicates that it is an important immunogen during disease. Production of the full-length recombinant Hia protein in *E. coli* appears to be toxic to the host, so a series of N-terminally truncated proteins was made as described in [copending] United States Patent [Application] No. 6,335,182 [09/268,347 filed March 16, 1999] and in PCT Patent Application No. PCT/CA00/00289 filed March 16, 2000, both assigned to the assignee hereof and the disclosures of which are incorporated herein by reference. The V38 rHia protein was chosen for further development as a vaccine, but it was found that the first 6 amino acids of this protein were deleted from a portion of the product during synthesis in *E. coli*, leading to a mixture of V38 rHia and S44 rHia. When an expression construct was developed to produce the S44 rHia, it was found that the N-terminus was stable, with only S44 rHia product being made. The rHia products appear as a doublet on SDS-PAGE when expressed alone. However, when co-expressed with H91A Hin47, the S44 rHia is produced as a single band, as described below."

Please replace the paragraph beginning at page 12, line 5, with the following rewritten paragraph:

"The *H. influenzae* Hia or Hsf proteins are demonstrated adhesins and as such are important vaccine candidates. The production of recombinant *H. influenzae* Hia proteins from *E. coli* has been described in the aforementioned US

Patent [Application] No. 6,335,182 [09/268,347]. The full-length proteins were expressed at very low levels and were apparently toxic to *E. coli*. A series of truncated rHia proteins was made, which were sequentially deleted at the N-terminus. The V38 rHia protein was produced as "soft" inclusion bodies and was purified, as described in the aforementioned US Patent [Application] No. 6,335,182 [09/268,347]. When the V38 rHia protein was co-produced with mature H91A Hin47, its solubility was increased. This led to an improved recovery during protein purification, and represents a novel use of mature H91A Hin47 (Figure 8). When analysed by SDS-PAGE, the V38 rHia protein was apparently produced as two doublets, whether or not it was co-produced with mature H91A Hin47 (Fig 6)."

Please replace the paragraph beginning at page 16, line 31, with the following rewritten paragraph:

"Plasmid DS-1872-2-2 is a pBR328-based vector containing a 2.2 kb *EcoR* I *T7 H91A hin47* gene cassette (Figure 5). Plasmid BK-96-2-11 is a pBR328-based vector that contains a *T7 V38 hia* gene cassette, the *E. coli cer* gene, and a kanamycin resistance gene; and this plasmid has been described in the aforementioned US Patent [Application] No. 6,335,182 [09/268,347]. BK-96-2-11 was linearized by digestion with *EcoR* I, dephosphorylated, and the *EcoR* I *T7 H91A hin47* gene fragment inserted, to generate plasmid DS-2342-2-2, that co-expresses the *H91A hin47* and *V38 hia* genes. This plasmid thus contains tandem *T7 H91A hin47* and *T7 V38 hia* genes in the same orientation. Plasmid DNA was introduced into electrocompetent *E. coli* BL21(DE3) cells using a BioRad electroporator, and recombinant *E. coli* strain DS-2350-3-1 was grown for protein analysis, as described in the following Example."

In the Claims:

Please cancel claims 14 to 43.

Please amend claims 4, 9, 12 and 13 as follows:

4. (Amended) The vector of claim 1 which is [plasmid vector having the identifying characteristics of] plasmid JB-3120-2 as seen in Figure 1A.

9. (Amended) The vector of claim 5 wherein said at least one additional nucleic acid molecule encodes a Hia [or Hsf] protein of a strain of *Haemophilus influenzae*.

12. (Amended) The vector of claim 11 which is [a plasmid vector having the identifying characteristics of] plasmid DS-2342-2-2 as seen in Figure 5.

13. (Amended) The vector of claim 11 which is [a plasmid vector having the identifying characteristics of] plasmid JB-3145-1 seen in Figure 10.

In the Drawings:

Please cancel Figures 17A, 18A and 19A and substitute therefor the new Figures of drawings enclosed.